

TITLE

Impact of the Nordic hamstring and hip extension exercises on hamstring architecture and morphology: implications for injury prevention

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ABSTRACT

The architectural and morphological adaptations of the hamstrings in response to training with different exercises have not been explored. **PURPOSE:** To evaluate changes in biceps femoris long head (BF_{LH}) fascicle length and hamstring muscle size following 10-weeks of Nordic hamstring exercise (NHE) or hip extension (HE) training. **METHODS:** Thirty recreationally active male athletes (age, 22.0 ± 3.6 years, height, 180.4 ± 7 cm, weight, 80.8 ± 11.1 kg) were allocated to one of three groups: 1) HE training (n=10), NHE training (n=10), or no training (CON) (n=10). BF_{LH} fascicle length was assessed before, during (Week 5) and after the intervention with 2D-ultrasound. Hamstring muscle size was determined before and after training via magnetic resonance imaging. **RESULTS:** Compared to *baseline*, BF_{LH} fascicles were lengthened in the NHE and HE groups at *mid-* ($d = 1.12 - 1.39$, $p < 0.001$) and *post-training* ($d = 1.77 - 2.17$, $p < 0.001$) and these changes did not differ significantly ($d = 0.49 - 0.80$, $p = 0.279 - 0.976$). BF_{LH} volume increased more for the HE than the NHE ($d = 1.03$, $p = 0.037$) and CON ($d = 2.24$, $p < 0.001$) groups. Compared to the CON group, both exercises induced significant increases in semitendinosus volume ($d = 2.16 - 2.50$, ≤ 0.002) and these increases were not significantly different ($d = 0.69$, $p = 0.239$). **CONCLUSION:** NHE and HE training both stimulate significant increases in BF_{LH} fascicle length, however, HE training may be more effective for promoting hypertrophy in the BF_{LH}.

What are the new findings?

- Hip extension and Nordic hamstring exercise training both promote the elongation of biceps femoris long head fascicles, and stimulate improvements in eccentric knee flexor strength.
- Hip extension training promotes more hypertrophy in the biceps femoris long head and semimembranosus than the Nordic hamstring exercise, which preferentially develops the semitendinosus and the short head of biceps femoris.

INTRODUCTION

Hamstring ‘tears’ are endemic in sports involving high-speed running and upwards of 80% of these injuries involve the biceps femoris long head (BF_{LH}). [1-4] Hamstring strains represent the most common injury in athletics, [5] Australian Rules football, [6 7] and soccer [8] and as many as 30% reoccur within 12 months. [9] These findings highlight the need for improved hamstring injury prevention programs while also suggesting the possibility that these programs should specifically target the BF_{LH}.

There has been significant interest in exploring the patterns of muscle activity in hamstring exercises, [10-15] however there is no research examining the architectural and morphological adaptations of these muscles to different exercise interventions. The Nordic hamstring exercise (NHE) has proven effective in increasing eccentric knee flexor strength [16] and reducing hamstring injuries [17-19] in soccer, although there is disagreement in the literature as to which hamstring muscles are most active during this exercise [10 14 15 20]. We have previously reported that the NHE preferentially activates the semitendinosus (ST), [10 15] however, we have also observed high levels of BF_{LH} activity in this exercise [15] which suggests that it may still provide a powerful stimulus for adaptation within this most commonly injured muscle. [1-4] Eccentric exercise has been proposed to increase muscle fascicle lengths via sarcomerogenesis [21 22] and Timmins and colleagues [23] have recently observed such an adaptation after eccentric knee flexor training on an isokinetic dynamometer while also noting that concentric training caused fascicle shortening despite occurring at long muscle lengths. Furthermore, we have recently reported that soccer players with shorter BF_{LH} fascicles (<10.56cm) were at fourfold greater risk of hamstring strain injury than players with longer fascicles. [23] Given the effectiveness of the predominantly eccentric NHE in hamstring injury prevention and rehabilitation, [17-19] it is of interest to

examine the impact of this and alternative exercises on BF_{LH} fascicle lengths and morphology.

We have recently observed that the 45° hip extension (HE) exercise resulted in more uniform activation of the two-joint hamstrings and greater BF_{LH} activity than the NHE[15]. HE exercises are also performed at longer hamstring muscle lengths than the NHE and it has been suggested that this may make them more effective in hamstring injury prevention than the NHE.[24] However, HE and most other hamstring exercises are typically performed with both eccentric and concentric phases and it remains to be seen how the combination of contraction modes will affect fascicle length by comparison with an almost purely eccentric exercise like the NHE. Nevertheless, the greater activation of BF_{LH} during HE[10 15] may provide a greater stimulus for hypertrophy, which might have implications for rehabilitation practices given observations of persistent atrophy in this muscle following injury.[25]

The primary purpose of this study was to evaluate changes in BF_{LH} architecture and hamstring muscle volume and anatomical cross-sectional area (ACSA) following 10-week resistance training programs consisting exclusively of NHE or HE training. We tested the hypotheses that 1) HE training would stimulate greater increases in BF_{LH} fascicle length than the NHE, on the basis of the suggestion that the ‘elongation stress’ in hamstring exercises may be an important factor in triggering this adaptation[24]; 2) HE training would promote more BF_{LH} hypertrophy than the NHE; and 3) the NHE would result in more hypertrophy of the ST muscle than the HE exercise.

METHODS

Participants

Thirty recreationally active males (age, 22.0 ± 3.6 years, height, 180.4 ± 7 cm, weight, 80.8 ± 11.1 kg) provided written informed consent to participate in this study. Participants were free from soft tissue and orthopaedic injuries to the trunk, hips and lower limbs and had no known history of hamstring strain, anterior cruciate ligament or other traumatic knee injury. Before enrolment in the study, all participants completed a cardiovascular screening questionnaire and a standard MRI questionnaire to ensure it was safe for them to enter the magnetic field. This study was approved by the Queensland University of Technology Human Research Ethics Committee and the University of Queensland Medical Research Ethics Committee.

Study design

This longitudinal training study was conducted between April and June, 2015. Approximately one week before the intervention commenced, participants underwent MR and 2D ultrasound imaging of their posterior thighs to determine hamstring muscle size and BF_{LH} architecture, respectively. After scanning, all participants were familiarised with the NHE and 45° HE exercise and subsequently underwent strength assessments on each exercise. After all of the pre-training assessments had been completed, participants were allocated to one of three groups: NHE, HE or control (CON). Allocation of participants to groups was performed on the basis of *baseline* BF_{LH} fascicle lengths to ensure that groups did not differ in this parameter prior to commencement of the study. Of the three participants with the longest fascicles, the first (with the longest fascicles) was allocated randomly to one of the three groups and then the second was allocated at random to one of the remaining two groups and the third allocated to the remaining group. This process was repeated for the participants with

the 4th to 6th longest fascicles, the 7th to 9th longest fascicles and so forth until each group had 10 participants. The NHE and HE groups completed a 10-week progressive strength training program consisting exclusively of their allocated exercise (Table 1). The CON group were advised to continue their regular physical activity levels but not to engage in any resistance training for the lower body. At the beginning of every training session, participants in both training groups reported their level of perceived soreness in the posterior thigh using a 1-10 numeric pain rating scale. All CON participants were required to report to the laboratory at least once per week. For all participants, BF_{LH} architecture was re-assessed 5 weeks into the intervention and within 5 days of the final training session. MRI scans were acquired for all participants <7 days after the final training session. Strength testing was conducted after all imaging had been completed.

Training intervention

Nordic hamstring exercise (NHE)

An illustration of the NHE can be found in Figure 1a (see also video supplement). Participants knelt on a padded board, with the ankles secured immediately superior to the lateral malleolus by individual ankle braces which were attached to uniaxial load cells. The ankle braces and load cells were secured to a pivot which allowed the force generated by the knee flexors to be measured through the long axis of the load cells. From the initial kneeling position with their ankles secured in yokes, arms on the chest and hips extended, participants lowered their bodies as slowly as possible to a prone position.[10] Participants performed only the lowering (eccentric) portion of the exercise and were instructed to use their arms and flex at the hips and knees to push back into the starting position so as to minimise concentric knee flexor activity. When participants developed sufficient strength to completely stop the movement in the final 10-20° of the range of motion, they were required to hold a weight

plate (range = 2.5kg to 20kg) to their chest (centred to the xiphoid process) to ensure the exercise was still of supramaximal intensity. Participants were provided with 3min of rest between each set.

Hip extension exercise (HE)

Participants were positioned in a 45° hip extension machine (BodySolid, IL, USA) with their trunk erect and hip joints extended and superior to the level of support pad (Figure 1b; see also video supplement). The ankle of the exercised limb was ‘hooked’ under an ankle pad and the unexercised limb was allowed to rest above its ankle restraint. Participants held one or more circular weight plate(s) to the chest (centred to the xiphoid process) and were instructed to flex their hip until they reached a point approximately 90° from the starting position. Once participants had reached this position they were instructed to return to the starting position by extending their hip, while keeping their trunk in a rigid neutral position throughout. Both limbs were trained in alternating fashion; after completing a set on one limb participants rested for 30s before training the opposite limb, and then recovered for 3min before the next set. The load held to the chest in week 1 represented 60-70% of the estimated 1-RM and was progressively increased throughout the training period whenever the prescribed repetitions and sets could be completed with appropriate technique (Table 2).

INSERT FIGURE 1

Hamstring training program

Participants in both intervention groups completed a progressive intensity training program consisting of 20 supervised exercise sessions (2 per week) over the 10 week period (Tables 1

& 2). Each session was followed by at least 48 hours of recovery and participants were prohibited from engaging in any other resistance training for the lower body. The training program was based on the approximate loads, repetitions and sets employed in previous interventions using the NHE,[16-18] although the volume (number of repetitions) was reduced in the final two weeks to accommodate increases in exercise intensity. All sessions were conducted in the same laboratory, employed the same exercise equipment and were supervised by the same investigators (MNB and SJD) to ensure consistency of procedures.

Table 1. Training program variables for both the Nordic hamstring and hip extension training groups

Week	Frequency	Sets	Repetitions
1	2	2	6
2	2	3	6
3	2	4	8
4	2	4	10
5-8	2	5	8-10
9	2	6	6
10	2	5	5

Table 2. Application of progressive overload for both the Nordic hamstring and hip extension training groups

Week	Training Intensity (Load)	
	Nordic Hamstring exercise	Hip extension exercise
1	Load was added to the chest in increments of 2.5kg when participants developed sufficient strength to stop at the end of the range of motion.	60-70% of 1-RM
2		70-80% of 1-RM
3		All exercise was completed at maximal intensity of effort. Loads were progressively increased when
4		
5-8		

9		desired repetitions and sets were
10		achieved.

Strength assessments

Before and <7 days after the intervention, all participants underwent an assessment of their maximal eccentric knee-flexor strength during three repetitions of the NHE, and their 3-repetition maximum (3-RM) strength on the 45° hip extension machine. All strength tests were conducted by the same investigators (MNB, SJD and AJS) with tests completed at approximately the same time of day before and after the intervention.

Nordic eccentric strength test

The assessment of eccentric knee flexor force using the NHE has been reported previously.[34 23 26] Participants completed a single warm-up set of 5 submaximal repetitions followed, 1 minute later, by a set of 3 maximal repetitions of the bilateral NHE. Eccentric strength was determined for each leg from the highest of 3 peak forces produced during the 3 repetitions of the NHE and was reported in absolute terms (N).

Hip extension strength test

All strength assessments on the 45° hip extension machine were conducted unilaterally. Participants initially warmed up by performing 8-10 repetitions on each leg using body weight only. Subsequently, loads held to the chest were progressively increased until investigators determined the maximal load that could be lifted three times. At least 2min of rest was provided between sets.

BF_{LH} architecture assessment

207 BF_{LH} fascicle length was determined from ultrasound images taken along the longitudinal
208 axis of the muscle belly utilising a two-dimensional, B-mode ultrasound (frequency, 12Mhz;
209 depth, 8cm; field of view, 14 x 47mm) (GE Healthcare Vivid-i, Wauwatosa, U.S.A).
210 Participants were positioned prone on a plinth with their hips in neutral and knees fully
211 extended, while images were acquired from a point midway between the ischial tuberosity
212 and the knee joint fold, parallel to the presumed orientation of BF_{LH} fascicles. After the
213 scanning site was determined, the distance of the site from various anatomical landmarks
214 were recorded to ensure its reproducibility for future testing sessions. These landmarks
215 included the ischial tuberosity, head of the fibula and the posterior knee joint fold at the mid-
216 point between BF and ST tendon. On subsequent visits the scanning site was determined and
217 marked on the skin and then confirmed by replicated landmark distance measures. Images
218 were obtained from both limbs following at least five minutes of inactivity. To gather
219 ultrasound images, the linear array ultrasound probe, with a layer of conductive gel was
220 placed on the skin over the scanning site, aligned longitudinally and perpendicular to the
221 posterior thigh. Care was taken to ensure minimal pressure was placed on the skin by the
222 probe as this may influence the accuracy of the measures.[27] The orientation of the probe
223 was manipulated slightly by the sonographer (RGT) if the superficial and intermediate
224 aponeuroses were not parallel.

225 Ultrasound images were analysed using MicroDicom software (Version 0.7.8, Bulgaria). For
226 each image, 6 points were digitised as described by Blazeovich and colleagues.[28] Following
227 the digitising process, muscle thickness was defined as the distance between the superficial
228 and intermediate aponeuroses of the BF_{LH}. A fascicle of interest was outlined and marked on
229 the image (Figure 2). Fascicle length was determined as the length of the outlined fascicle
230 between aponeuroses and was reported in absolute terms (cm). As the entire fascicles were

not visible in the probe's field of view, their lengths were estimated using the following equation:[28 29]

$$FL = \sin(AA + 90^\circ) \times MT / \sin(180^\circ - (AA + 180^\circ - PA)).$$

Where FL =fascicle length, AA =aponeurosis angle, MT =muscle thickness and PA =pennation angle.

All images were collected and analysed by the same investigator (RGT) who was blinded to training group allocation. The assessment of BF_{LH} architecture using the aforementioned procedures by this investigator (RGT) is highly reliable (intraclass correlations >0.90).[30]

INSERT FIGURE 2

Muscle volumes and anatomical cross-sectional area assessment All MRI scans were performed using a 3-Tesla (Siemens TrioTim, Germany) imaging system with a spinal coil. The participant was positioned supine in the magnet bore with the knees fully extended and hips in neutral, and straps were placed around both limbs to prevent any undesired movement. Contiguous T1-weighted axial MR images (transverse relaxation time: 750ms; echo time: 12ms; field of view: 400mm; slice thickness: 10mm; interslice distance: 0mm) were taken of both limbs beginning at the iliac crest and finishing distal to the tibial condyles. A localiser adjustment (20s) was applied prior to the acquisition of T1-weighted images to standardise the field of view. In addition, to minimise any inhomogeneity in MR images caused by dielectric resonances at 3T, a post-processing (B1) filter was applied to all scans.[31] The total scan duration was 3min 39sec.

Muscle volumes and anatomical cross-sectional areas (ACSAs) of the BF_{LH} and short head (BF_{SH}), semitendinosus (ST) and semimembranosus (SM) muscles were determined for both

limbs using manual segmentation. Muscle boundaries were identified and traced on each image in which the desired structure was present using image analysis software (Sante Dicom Viewer and Editor, Cornell University) (Figure 3). Volumes were determined for each muscle by multiplying the summed CSAs (from all the slices containing the muscle of interest) by the slice thickness.[25] ACSA was determined by locating the 10mm slice with the greatest CSA and averaging this along with the two slices immediately cranial and caudal (five slices). All traces (pre- and post-training) were completed by the same investigator (MNB) who was blinded to participant identity and training group in all post-testing.

INSERT FIGURE 3

Statistical analysis

All statistical analyses were performed using SPSS version 22.0.0.1 (IBM Corporation, Chicago, IL). Repeated measures split plot ANOVAs were used to determine training-induced changes in BF_{LH} architecture, hamstring muscle volumes and ACSA, strength, and ratings of perceived soreness, for each group. For the analysis of BF_{LH} fascicle length, the within-subject variable was *time (baseline, mid-training, and post-training)* and the between-subject variable was *group* (HE, NHE, CON). Because BF_{LH} architecture did not differ between limbs (dominant vs non-dominant) at any time point ($p>0.05$), the left and right limbs were averaged to provide a single value for each participant. To determine differences in the percentage change in hamstring muscle volume and ACSA between groups, the within-subject variable was *muscle* (BF_{LH}, BF_{SH}, ST, and SM) and the between-subject variable was *group* (HE, NHE, CON). To explore changes in Nordic and 45° hip extension strength the within-subject variable was *time (baseline and post-training)* and the between-subject variable was *group* (HE, NHE, CON). Lastly, to determine if ratings of perceived soreness

changed over time, or differed between training groups, within-subject variable was *time* (weeks 1-10) and the between-subject variable was *group* (HE, NHE, CON) For all analyses, when a significant main effect was detected, post hoc independent t tests with Bonferroni corrections were used to determine which comparisons differed. For all analyses, the mean differences were reported with their 95% confidence intervals (CIs), and where appropriate, Cohen's *d* was reported as a measure of the effect size.

Sample size

A priori sample size estimates were based on anticipated differences in BF_{LH} fascicle length following the training intervention. A sample size of 10 in each group was calculated to provide sufficient statistical power (80%) to detect an effect size of 1.0 for the difference in fascicle length changes between training groups, with $p < 0.05$.

RESULTS

No significant differences were observed in age, height or body mass between the three groups ($p > 0.05$) (Table 3). Compliance rates were excellent for both training groups (HE: 100%; NHE: 99.5%).

Table 3. Participant characteristics

Group	Age (years)	Height (cm)	Mass (kg)
HE	23.1±4.1	180±6.3	81.6±9.7
NHE	21.6±3.2	182.8±8.7	85.0±10.9
CON	21.3±3.7	178.5±5.4	75.9±11.8

Biceps femoris long head (BF_{LH}) fascicle length

Between-group comparisons

A significant *group × time* interaction was observed for fascicle length during the training period ($p < 0.001$) (Figure 4). No significant differences were observed between training groups at either *baseline* ($d = 0.15$), *mid-* ($d = 0.49$) or *post-training* points ($d = 0.80$) (all $p > 0.05$). However, the NHE group displayed significantly longer fascicles than the CON group at *mid-* (mean difference = 1.50cm, 95% CI = 0.58 to 2.41cm, $d = 1.64$, $p = 0.001$) and *post-training* (mean difference = 2.40cm, 95% CI = 1.28 to 3.53cm, $d = 2.19$, $p < 0.001$). Similarly, the HE group exhibited significantly longer fascicles than the CON group at *mid-* (mean difference = 1.14cm, 95% CI = 0.22 to 2.05cm, $d = 1.52$, $p = 0.011$) and *post-training* (mean difference = 1.63cm, 95% CI = 0.51 to 2.76cm, $d = 1.84$, $p = 0.003$).

Within-group comparisons

Post hoc analyses revealed that BF_{LH} fascicle length increased significantly from *baseline* in the NHE group at *mid-* (mean difference = 1.23cm, 95% CI = 0.84 to 1.63cm, $d = 1.39$, $p < 0.001$) and *post-training* (mean difference = 2.22cm, 95% CI = 1.74 to 2.69cm, $d = 2.17$, $p < 0.001$). The HE group also displayed significantly lengthened fascicles at *mid-* (mean difference = 0.75cm, 95% CI = 0.35 to 1.15cm, $d = 1.12$, $p < 0.001$) and *post-training* (mean difference = 1.33cm, 95% CI = -0.86 to 1.80cm, $d = 1.77$, $p < 0.001$). However, the CON group remained unchanged relative to *baseline* values at all time points ($p > 0.05$, $d = 0.20 - 0.31$).

INSERT FIGURE 4

Hamstring muscle volumes

Between-group comparisons

A significant main effect was detected for the *muscle x group* interaction for hamstring muscle volume changes ($p < 0.001$) (Figure 5). BF_{LH} volume increased significantly more in the HE than the NHE (mean difference = 6.72%, 95% CI = 0.32 to 13.11%, $d = 1.03$, $p = 0.037$) and CON groups (mean difference = 12.10%, 95% CI = 5.71 to 18.50%, $d = 2.24$, $p < 0.001$), and a smaller nonsignificant difference was observed between the NHE and CON groups (mean difference = 5.39%, 95% CI = -1.01 to 11.78%, $d = 1.13$, $p = 0.122$) (Figure 5). BF_{SH} volume increased more in the HE (mean difference = 8.51%, 95% CI = 0.17 to 16.85%, $d = 1.49$, $p = 0.044$) and NHE groups (mean difference = 15.29%, 95% CI = 6.95 to 23.63%, $d = 2.09$, $p < 0.001$) than in the CON group. Both the NHE (mean difference = 21.21%, 95% CI = 11.55 to 30.88%, $d = 2.50$, $p < 0.001$) and HE (mean difference = 14.32%, 95% CI = 4.65 to 23.98%, $d = 2.16$, $p = 0.002$) training groups exhibited a greater

increase in ST volume than the CON group. However, no significant difference in ST volume change was noted between NHE and HE groups (mean difference = 6.90%, 95% CI = -2.77 to 16.56%, $d = 0.69$, $p = 0.239$). The percentage change in volume for the SM was significantly greater for the HE group than for CON (mean difference = 8.95%, 95% CI = 2.21 to 15.69%, $d = 1.57$, $p = 0.007$), while no difference was observed between the NHE and CON group changes (mean difference = 3.38%, 95% CI = -3.36 to 10.12%, $d = 0.68$, $p = 0.636$) for this muscle.

Within-group comparisons

HE training stimulated a greater increase in volume for the ST than the BF_{SH} (mean difference = 5.61%, 95% CI = 1.12% to 10.10%, $d = 0.71$, $p = 0.009$). No other significant between-muscle differences were noted for volume changes after HE training ($p=0.054 - 0.999$ for all pairwise comparisons) or in the CON group ($p > 1.000$). After NHE training, ST volume increased more than BF_{LH} (mean difference = 15.28%, 95% CI = 10.69 to 19.87%, $d = 3.54$, $p<0.001$) and SM (mean difference = 16.06%, 95% CI = 10.96 to 21.16%, $d = 3.53$, $p<0.001$). Similarly, in the NHE group the percentage change in volume was greater for the BF_{SH} than the BF_{LH} (mean difference = 9.56%, 95% CI = 4.30 to 14.80%, $d = 1.18$, $p < 0.001$) and SM (mean difference = 10.33%, 95% CI = 5.33 to 15.34%, $d = 1.26$, $p < 0.001$).

INSERT FIGURE 5

Hamstring muscle anatomical cross-sectional area (ACSA)

Between-group comparisons

A significant main effect was detected for the *muscle x group* interaction ($p < 0.001$) (Figure 6). The percentage change in BF_{LH} ACSA was greater in the HE training group than in the

NHE (mean difference = 5.24%, 95% CI = 0.061 to 10.41, $d = 0.98$, $p = 0.047$) and CON groups (mean difference = 8.90%, 95% CI = 3.73 to 14.07%, $d = 1.94$, $p < 0.001$), while no difference was observed between the NHE and CON groups (mean difference = 3.67%, 95% CI = -1.51 to 8.84%, $d = 1.07$, $p = 0.245$) (Figure 6). BF_{SH} ACSA increased significantly more in the NHE than the CON group (mean difference = 13.26%, 95% CI = 4.98 to 21.54%, $d = 1.97$, $p = 0.001$), while no difference was observed between changes exhibited by the HE and CON groups for this muscle (mean difference = 5.69%, 95% CI = -2.59 to 0.70%, $d = 0.90$, $p = 0.273$). The percentage change in ST ACSA was significantly greater in the NHE (mean difference = 17.60%, 95% CI = 7.60 to 27.61%, $d = 2.17$, $p < 0.001$) and HE (mean difference = 15.16%, 95% CI = 5.15 to 25.17%, $d = 1.95$, $p = 0.002$) groups than the CON group, however no significant difference was noted between changes in the NHE and HE groups (mean difference = 2.4%, 95% CI = -7.57 to 12.45%, $d = 0.24$, $p = 1.000$). The percentage increase in SM ACSA was greater in the HE than the CON group (mean difference = 7.19%, 95% CI = 1.21 to 13.18%, $d = 1.34$, $p = 0.015$), but was not significantly greater in NHE than CON (mean difference = 2.02%, 95% CI = -3.97 to 8.01%, $d = 0.49$, $p = 1.000$). No significant difference in SM ACSA change was noted between the HE and NHE groups (main difference = 5.17%, 95% CI = -8.2 to 11.16%, $d = 0.85$, $p = 0.109$).

Within-group comparisons

After HE training, the change in ACSA observed for the ST was significantly greater than the BF_{LH} (mean difference = 6.46, 95% CI = 0.84 to 12.10%, $d = 0.78$, $p = 0.017$), BF_{SH} (mean difference = 9.98%, 95% CI = 4.25 to 15.71%, $d = 1.09$, $p < 0.001$) and SM (mean difference = 6.73%, 95% CI = 1.54 to 11.92%, $d = 0.78$, $p = 0.006$). No other significant pairwise between-muscle differences in ACSA change were noted after HE training (all $p > 0.05$). After NHE training, the change in ACSA was greater for BF_{SH} than BF_{LH} (mean difference = 9.30%, 95% CI = 3.47 to 15.12%, $d = 1.34$, $p = 0.001$) and SM (mean difference = 9.50%,

95% CI = 4.92 to 14.08, $d = 1.33$, $p < 0.001$), while ST ACSA increased more than BF_{LH} (mean difference = 14.14%, 95% CI = 8.52 to 19.76%, $d = 1.76$, $p < 0.001$) and SM (mean difference = 14.35%, 95% CI = 9.15 to 19.54%, $d = 1.75$, $p < 0.001$).

INSERT FIGURE 6

Strength

Nordic eccentric strength test

A significant *group x time* interaction effect was observed for the Nordic eccentric strength test ($p < 0.001$) (Figure 7). Post hoc *t* tests demonstrated that the NHE (mean difference = 97.38N, 95% CI = 65.51 to 129.26N, $d = 2.36$, $p < 0.001$) and HE (mean difference = 110.47N, 95% CI = 76.87 to 144.07N, $d = 1.26$, $p < 0.001$) groups were significantly stronger at *post-training* compared to *baseline* while the CON group did not change (mean difference = 8.91N, 95% CI = -42.51 to 24.69N, $d = 0.14$, $p = 0.590$). No groups differed at *baseline* ($p > 0.461$), however, at *post-training* the NHE (mean difference = 123.436N, 95% CI = 39.93 to 206.93N, $d = 2.07$, $p = 0.003$) and HE (mean difference = 94.27N, 95% CI = 8.60 to 179.94N, $d = 1.14$, $p = 0.028$) groups were both significantly stronger than the CON group. No significant difference was observed between training groups at *post-training* (mean difference = 29.16N, 95% CI = -54.34 to 112.66N, $d = 0.41$, $p = 0.999$).

INSERT FIGURE 7

Hip extension strength test

A significant *group x time* interaction effect was also observed for 3-RM strength as assessed during the 45° HE strength test ($p < 0.001$) (Figure 8). Post hoc analyses demonstrated that the HE (mean difference = 41.00kg, 95% CI = 35.97 to 46.03kg, $d = 4.59$, $p < 0.001$) and NHE groups (mean difference = 26.00kg, 95% CI = 20.97 to 31.03kg, $d = 2.36$, $p < 0.001$) improved significantly from *baseline* whereas the CON group did not change (mean difference = 3.50kg, 95% CI = -1.53 to 8.53kg, $d = 0.33$, $p = 0.165$). No groups differed significantly at *baseline* ($p > 0.091$) however at *post-training*, both the HE (mean difference = 43.50kg, 95% CI = 30.93 to 56.07kg, $d = 4.21$, $p < 0.001$) and NHE groups (mean difference = 32.0kg, 95% CI = 19.43 to 44.57kg, $d = 2.66$, $p < 0.001$) were significantly stronger than CON. *Post-training*, no significant difference was observed between training groups (mean difference = 11.50kg, 95% CI = -1.07 to 24.07kg, $d = 1.09$, $p = 0.082$).

INSERT FIGURE 8

Perceived soreness

No significant *group x time* interaction effect ($p = 0.397$) was detected for ratings of perceived soreness throughout the intervention (Figure 9). The average soreness measures reported across the 10-week training period were 2.2 ± 0.4 (mean \pm SE) for the NHE group and 2.3 ± 0.5 for the HE group.

INSERT FIGURE 9

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DISCUSSION

This study is the first to explore the architectural and morphological adaptations of the hamstrings in response to different strength training exercises. These data suggest that both the HE and NHE stimulate significant increases in BF_{LH} fascicle length and, contrary to our hypothesis, that the longer muscle lengths encountered in the HE exercise do not result in greater lengthening of fascicles than are observed after NHE training. As hypothesised, HE training appears to elicit more hypertrophy in the BF_{LH} than does the NHE; while contrary to our hypothesis, the NHE was not significantly more effective at increasing ST volume or cross sectional area than the HE. Both exercises resulted in significant strength increases which were similarly evident in the NHE and HE strength tests.

Fascicle lengthening is one possible mechanism by which the NHE[17-19] and other eccentric or long length hamstring exercises[22] protect muscles from injury. We have recently shown, prospectively, that professional soccer players with fascicles $<10.56\text{cm}$ were ~4 times more likely to suffer a hamstring strain than athletes with longer fascicles and that the probability of injury was reduced by ~74% for every 0.5cm increase in fascicle length.[23] In the current study, participants increased their fascicle lengths from ~10.6cm prior to training, to 12.8 and 12.0cm in the NHE and HE groups, respectively, which would likely result in large reductions in hamstring injury risk.

Despite its success in reducing hamstring strain injuries, the adoption of the NHE in elite European soccer has been reported to be poor with only ~11% of Norwegian premier league and UEFA teams deemed to have adequately implemented the NHE programs that have proven effective in randomised controlled trials[17-19]. Some conditioning coaches and researchers[24] believe that the exercise does not challenge the hamstrings at sufficient lengths to optimise injury prevention benefits. However, this study shows, for the first time, that the limited excursion of the hamstrings during the NHE does not prevent the exercise

from increasing BF_{LH} fascicle length. Indeed, the exercise resulted in greater fascicle lengthening than the HE, although the current study lacked the statistical power to distinguish between the two. Together with observations that long length concentric hamstring training can shorten muscle fascicles,[33] the current findings are consistent with the possibility that the combination of concentric and eccentric contractions somewhat dampens the elongation of BF_{LH} fascicles. The advantage of the NHE may be its almost purely eccentric or eccentrically-biased nature. Further work is needed to clarify whether eccentrically-biased or purely eccentric HE exercise may yield greater improvements in BF_{LH} fascicle length than the combined concentric and eccentric contraction modes used in this investigation.

Observations of increased fascicle length following eccentric hamstring exercise are largely consistent with existing literature. For example, Potier and colleagues[32] reported a 34% increase in BF_{LH} fascicle length following eight weeks of eccentric leg curl exercise, while Timmins and colleagues[33] reported a 16% increase in BF_{LH} fascicle length after six weeks of eccentric training on an isokinetic dynamometer.[33] These adaptations most likely result from the addition of in-series sarcomeres, as has been shown to occur within the rat vastus intermedius muscle after five days of downhill running.[34] It has been proposed that this increase in serial sarcomeres accounts for both a rightward shift in a muscle's force-length relationship,[35] while also reducing its susceptibility to damage.[21 22] However, it is also at least theoretically possible that fascicle lengthening occurs as a result of increased tendon or aponeurotic stiffness[41] and further research is needed to clarify the precise mechanism(s) responsible for these architectural changes.

To the authors' knowledge, this is the first study to explore the morphological adaptations of the hamstrings to different strengthening exercises. These data suggest that the NHE and HE exercises induce heterogeneous patterns of hamstring muscle hypertrophy, with the former preferentially stimulating ST and BF_{SH} growth and the latter resulting in significantly more

487 hypertrophy of the BF_{LH} and more homogenous growth of all two-joint hamstring muscles.
488 We have previously noted transient T2 relaxation time changes after 50 repetitions of each of
489 these exercises that almost exactly fit this pattern,[15] so it appears that the acute changes
490 observed via functional MRI match quite well with the hypertrophic effects observed after 10
491 weeks of training. However, neither muscle volume nor ACSA have been identified as risk
492 factors for hamstring strain injury, so the exact significance of these findings is unknown.
493 Indeed, we have previously reported that BF_{LH} muscle thickness measured via ultrasound is
494 not a risk factor for hamstring injury in elite soccer.[23] Nevertheless, BF_{LH} muscle atrophy
495 has been noted as long as 5-23 months after injury in recreational athletes,[25] so unilateral
496 HE exercises may prove more beneficial than the NHE at redressing this deficit in
497 rehabilitation. Interestingly, reduced muscle volumes of the ST have been observed 12-72
498 months after anterior cruciate ligament injury[36] and the results of the current investigation
499 suggest that the NHE may be valuable in rehabilitation of this injury.

500 Hamstring strengthening is an important component of injury prevention strategies.[24 37 38]
501 Indeed, several large scale interventions employing the NHE have shown ~65% reductions in
502 hamstring strain injury rates in soccer [17-19] and recent prospective findings in elite
503 Australian football[3] and soccer[23] suggest that eccentric strength improvements like those
504 reported here and previously[16] are at least partly responsible for these protective benefits.
505 For example, elite athletes in these sports who generated less than 279N (Australian football)
506 and 337N (soccer) of knee flexor force at the ankles during the NHE strength test were ~4
507 times more likely to sustain hamstring injuries than stronger counterparts.[3 23] In this study,
508 our recreational level athletes were able to generate, on average, 460N and 431N after 10
509 weeks of NHE and HE training, respectively, making them substantially stronger than these
510 elite Australian football[3] and soccer players.[23] Significant improvements in 3-RM HE
511 strength were also observed for both training groups, which suggests that hamstring

strengthening, at least in recreationally trained athletes, is not highly specific to the chosen exercise. While the benefits of high levels of HE strength remain unclear from the perspective of injury prevention, the observed effects of HE training on BF_{LH} fascicle lengths and eccentric knee flexor strength suggest the potential for this exercise to reduce injury risk. Future intervention studies analogous to those employing the NHE previously,[17-19 39] are needed to clarify whether HE training is effective in reducing hamstring strain injuries, however, access to exercise equipment (ie., a 45° HE machine) may be a limiting factor in designing such studies. It is also noteworthy that strength improvements can be achieved with very modest levels of hamstring muscle soreness when training is appropriately structured and progressively overloaded. These observations are in agreement with Mjolsnes and colleagues[16] who have previously reported very limited muscle soreness with a gradual increase in NHE volume.

The authors acknowledge that there are some limitations associated with the current study. Firstly, muscle architecture was only assessed in the BF_{LH} and it may not be appropriate to generalise these findings to other knee flexors, given that each hamstring muscle displays unique architectural characteristics.[40] Further, the assessment of fascicle length using two-dimensional ultrasound requires some degree of estimation, because the entire length of the BF_{LH} fascicles are not visible in ultrasound images. While the estimation equation used in this study has been validated against cadaveric samples,[29] there is still the potential for error, and future studies employing extended field of view ultrasound methods may be needed to completely eliminate this. Lastly, all of the athletes in this study were recreational level males of a similar age, and it remains to be seen if these results are applicable to other populations. However, our participants were, on average, as strong as elite Australian football players[3] and stronger than professional soccer players[23] at the start of the study. Furthermore, our cohort displayed average fascicle lengths before training that were within

one standard deviation of the values reported in elite soccer players previously,[23] so it is unlikely that they were unrepresentative of higher-level athletes, in these parameters at least.

This is the first study to demonstrate that training with different exercises elicits unique architectural and morphological adaptations within the hamstring muscle group. We have provided evidence to suggest that both HE and NHE training are effective in lengthening BF_{LH} fascicles and that the greater excursion involved in the HE does not result in greater increases in fascicle length. However, HE training appears to be more effective for promoting hypertrophy in the commonly injured BF_{LH} than the NHE, which preferentially develops the ST and BF_{SH} muscles. HE and NHE had very similar effects on ST volume and cross-sectional area. These data may help to explain the mechanism(s) by which the NHE confers injury preventive benefits and also provide compelling evidence to warrant the further exploration of HE-oriented exercises in hamstring strain injury prevention protocols. Future prospective studies are needed to ascertain whether HE training interventions are effective in reducing the incidence of hamstring strain injury in sport and whether or not the combination of HE and NHE training is more effective than the NHE alone.

How might it impact upon clinical practice in the future?

- Hip extension and Nordic hamstring exercise training are both effective in lengthening biceps femoris long head fascicles, and in promoting improvements in eccentric knee flexor strength, which may significantly reduce the risk of hamstring strain injury
- Hip extension exercise may be more useful than the Nordic hamstring exercise for stimulating hypertrophy in the commonly injured biceps femoris long head

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CONTRIBUTORS

MB was the principle investigator and was involved with study design, recruitment, analysis and manuscript write up. SD, RT were involved in data collection. MW, DO, GK and TS were involved with the study design, analysis and manuscript preparation. AA was involved in MRI data acquisition. All authors had full access to all of the data (including statistical reports and tables) in the study and can take responsibility for the integrity of the data and the accuracy of the data analysis.

TRANSPARENCY DECLARATION

The lead author* (MB) affirms that this manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned (and, if relevant, registered) have been explained. * = The manuscript's guarantor.

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DATA SHARING

Consent was not obtained for data sharing but the presented data are anonymised and risk of identification is low.

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COMPETING INTERESTS

None declared. All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare that (1) the Queensland Academy of Sport's Centre of Excellence for Applied Sports Science Research funded this study; (2) MB, SD, RT, MW, DO, GK, AA and TS have no relationships with companies that might have an interest in the submitted work in the previous 3 years; (3) their spouses, partners, or children have no financial relationships that may be relevant to the submitted work; and (4) MB, SD, RT, MW, DO, GK, AA and TS have no non-financial interests that may be relevant to the submitted work.

ETHICAL CLEARANCE

All participants provided written, informed consent for this study, which was approved by the Queensland University of Technology Human Research Ethics Committee and the University of Queensland Medical Research Ethics Committee.

Figure legends

Figure 1. (a) The Nordic hamstring exercise (NHE) and **(b)** the 45° hip extension (HE) exercise, progressive from left to right.

Figure 2. A two-dimensional ultrasound image of the biceps femoris long head (BF_{LH}), taken along the longitudinal axis of the posterior thigh. From these images, it is possible to determine the superficial and intermediate aponeuroses, muscle thickness, and angle of the fascicle in relation to the aponeurosis. Estimates of fascicle length can then be made via trigonometry using muscle thickness and pennation angle.

Figure 3. T1-weighted image (transverse relaxation time = 750ms; echo time = 12ms, slice thickness = 10mm), depicting the regions of interest for each hamstring muscle. The *right* side of the image corresponds to the participant's *left* side as per radiology convention. BF_{LH}, biceps femoris long head; BF_{SH}, biceps femoris short head; ST, semitendinosus; SM, semimembranosus.

Figure 4. Biceps femoris long head (BF_{LH}) fascicle lengths before (*baseline*), during (*mid-training*) and after (*post-training*) the intervention period for the hip extension (HE), Nordic hamstring exercise (NHE) and control (CON) groups. Fascicle length is expressed in absolute terms (cm) with error bars depicting standard error (SE). * indicates $p < 0.05$ compared to *baseline* (week 0). ** signifies $p < 0.001$ compared to *baseline*. # indicates $p < 0.05$ compared to the control group.

Figure 5. Percentage change in volume (cm³) for each hamstring muscle after the intervention period for the hip extension (HE), Nordic hamstring exercise (NHE) and control (CON) groups. Values are expressed as a mean percentage change compared to the values at *baseline* with error bars representing standard error (SE). For all pairwise comparisons

between groups, * indicates $p<0.05$ and ** signifies that $p<0.001$. BF_{LH}, biceps femoris long head; BF_{SH}, biceps femoris short head; ST, semitendinosus; SM, semimembranosus.

Figure 6. Percentage change in anatomical cross sectional area (ACSA) (cm²) for each hamstring muscle after the intervention period for the hip extension (HE), Nordic hamstring exercise (NHE) and control (CON) groups. Values are expressed as a mean percentage change compared to the values at *baseline* with error bars representing standard error (SE). For all pairwise comparisons between groups, * indicates $p<0.05$ and ** signifies that $p<0.001$. BF_{LH}, biceps femoris long head; BF_{SH}, biceps femoris short head; ST, semitendinosus; SM, semimembranosus.

Figure 7. Eccentric knee flexor force measured during the Nordic strength test before (*baseline*) and after (*post-training*) the intervention period for the hip extension (HE), Nordic hamstring exercise (NHE) and control (CON) groups. Force is reported in absolute terms (N) with error bars depicting standard error (SE). * indicates $p<0.001$ compared to *baseline* (week 0). # signifies $p<0.05$ compared to the control group.

Figure 8. Hip extension three-repetition maximum (3RM) before (*baseline*) and after (*post-training*) the intervention period for the hip extension (HE), Nordic hamstring exercise (NHE) and control (CON) groups. Force is reported in absolute terms (kg) with error bars depicting standard error (SE). ** indicates $p<0.001$ compared to *baseline* (week 0). # signifies $p<0.001$ compared to the control group.

Figure 9. Mean (\pm standard error) weekly soreness measured using a numeric pain rating scale (1-10) at the beginning of each training session for the hip extension (HE) and Nordic hamstring exercise (NHE) groups.

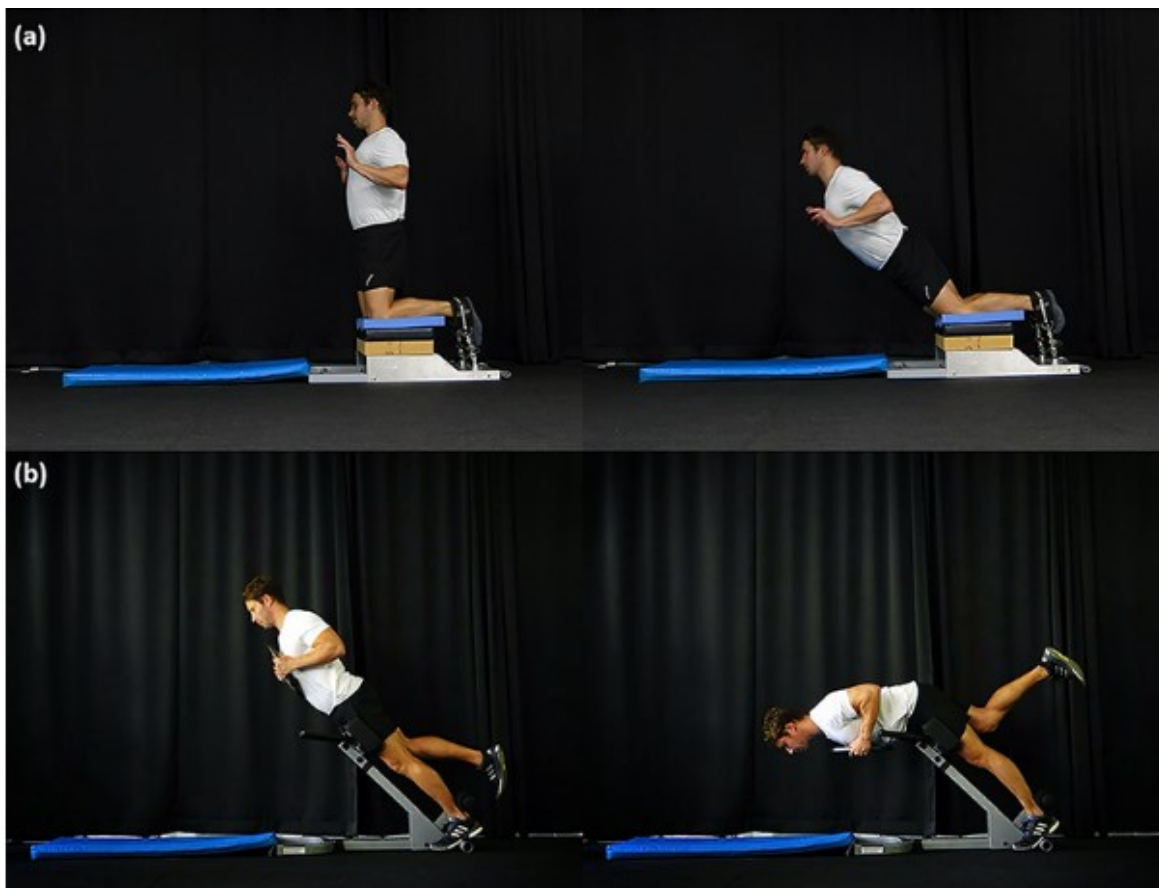
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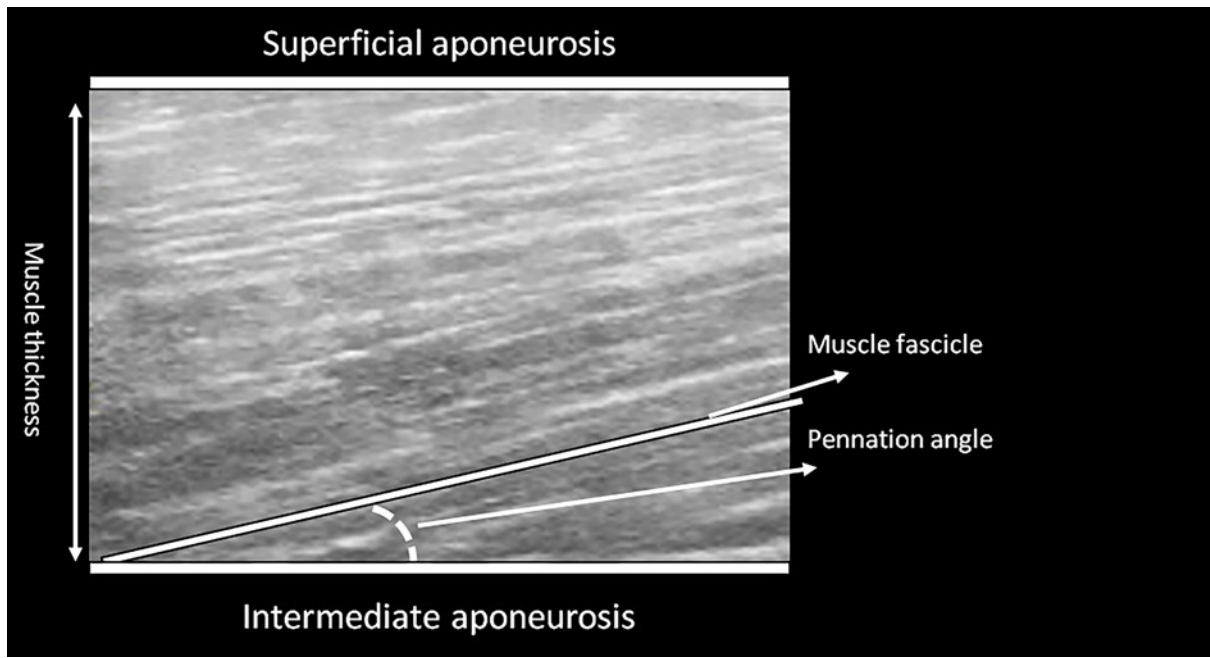
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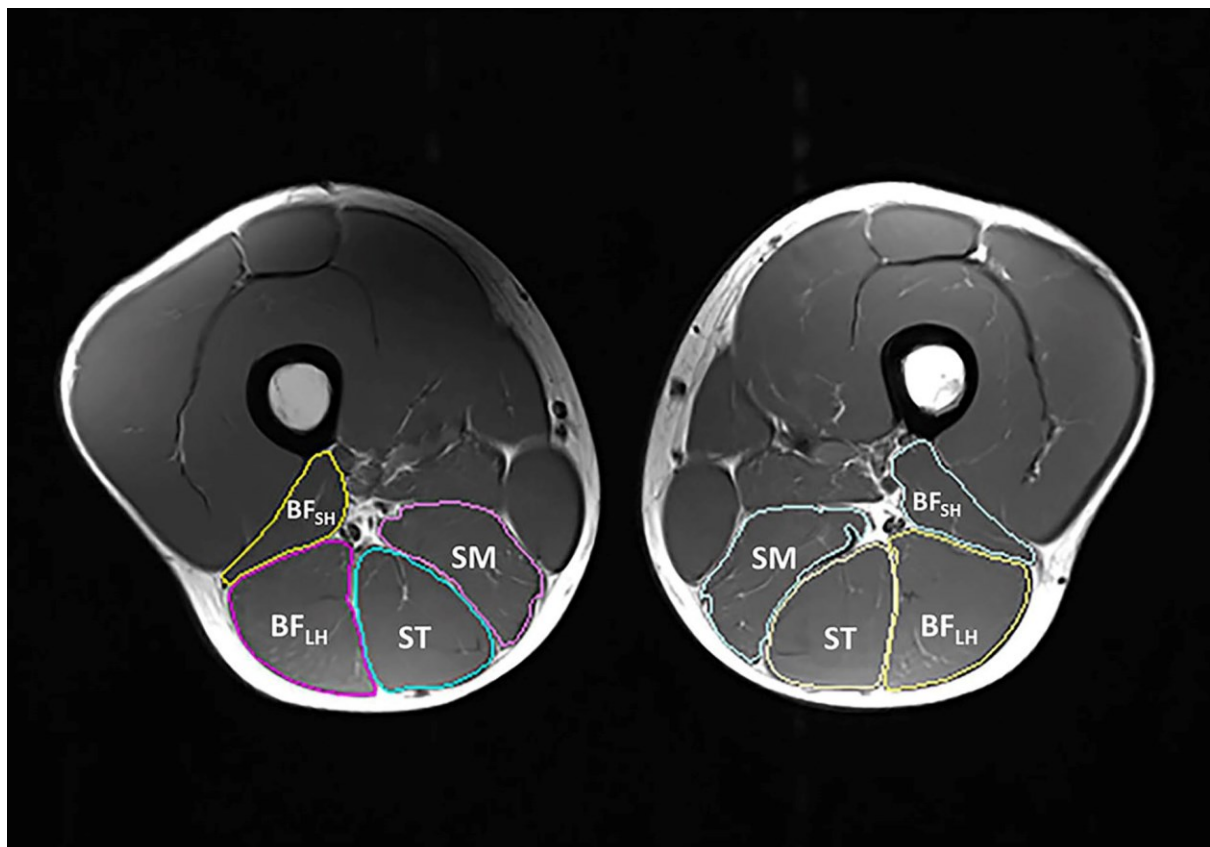




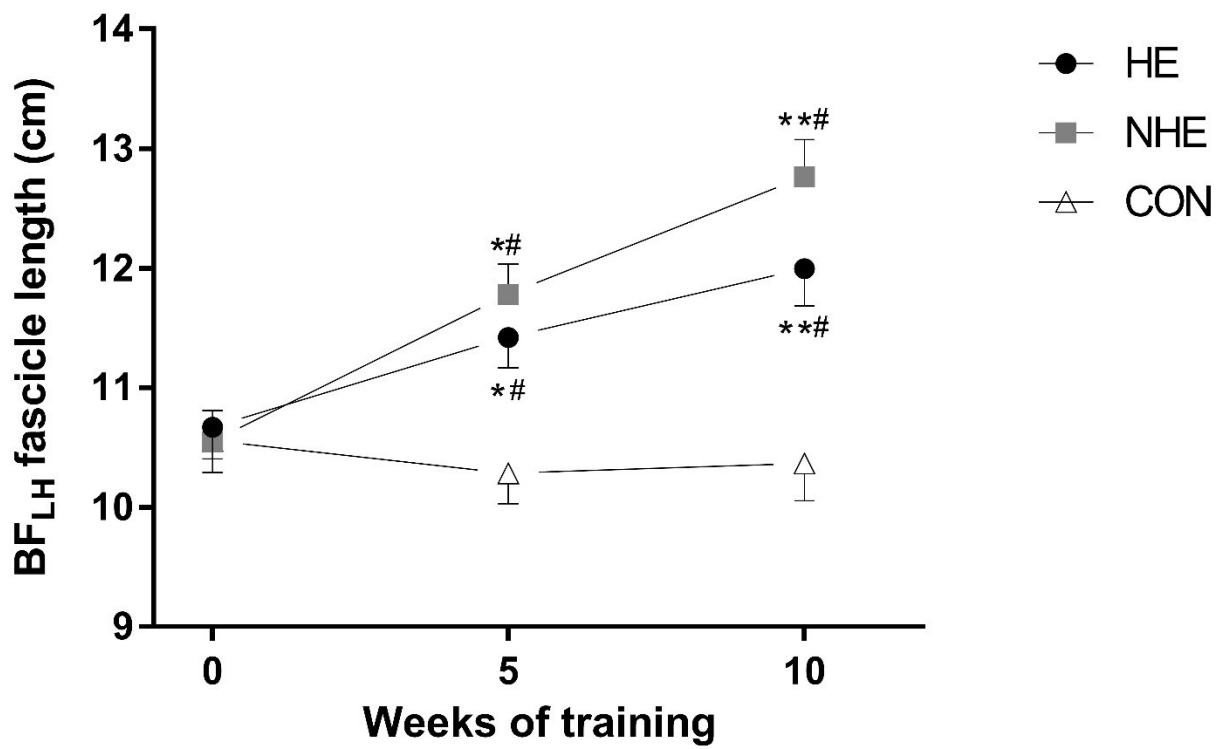
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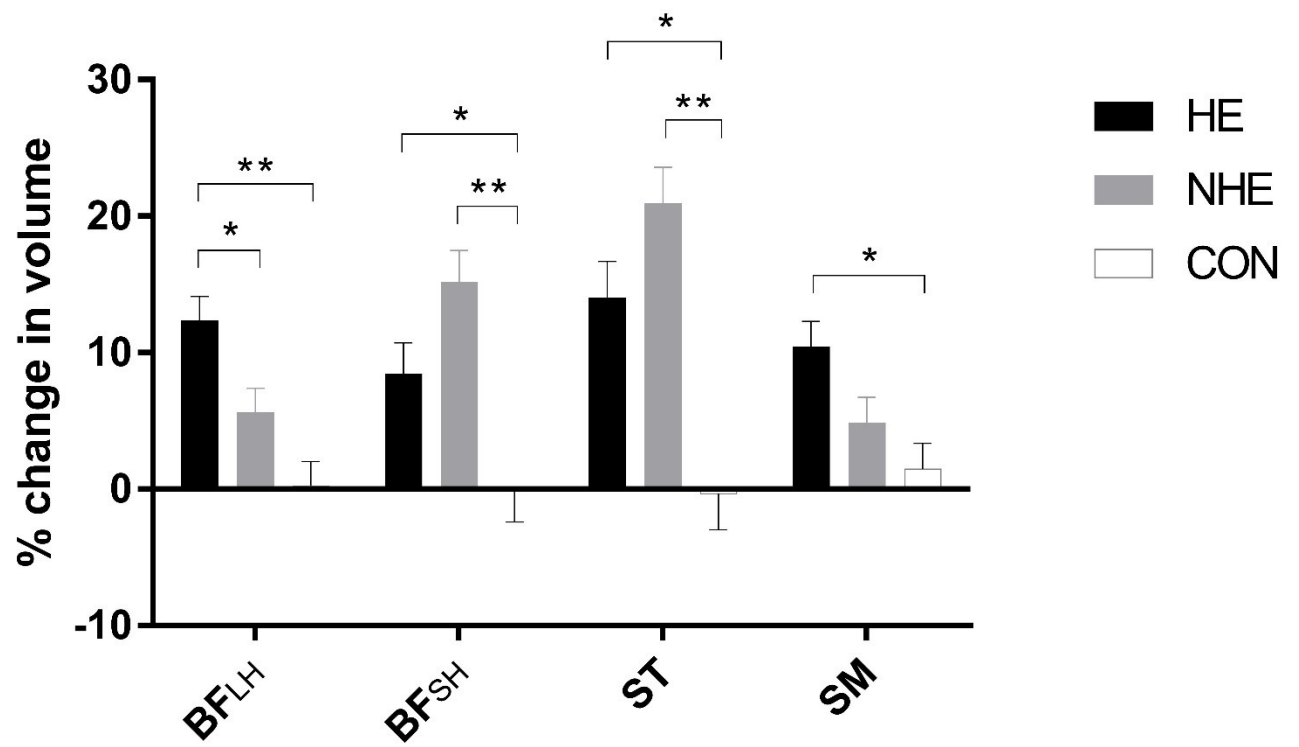
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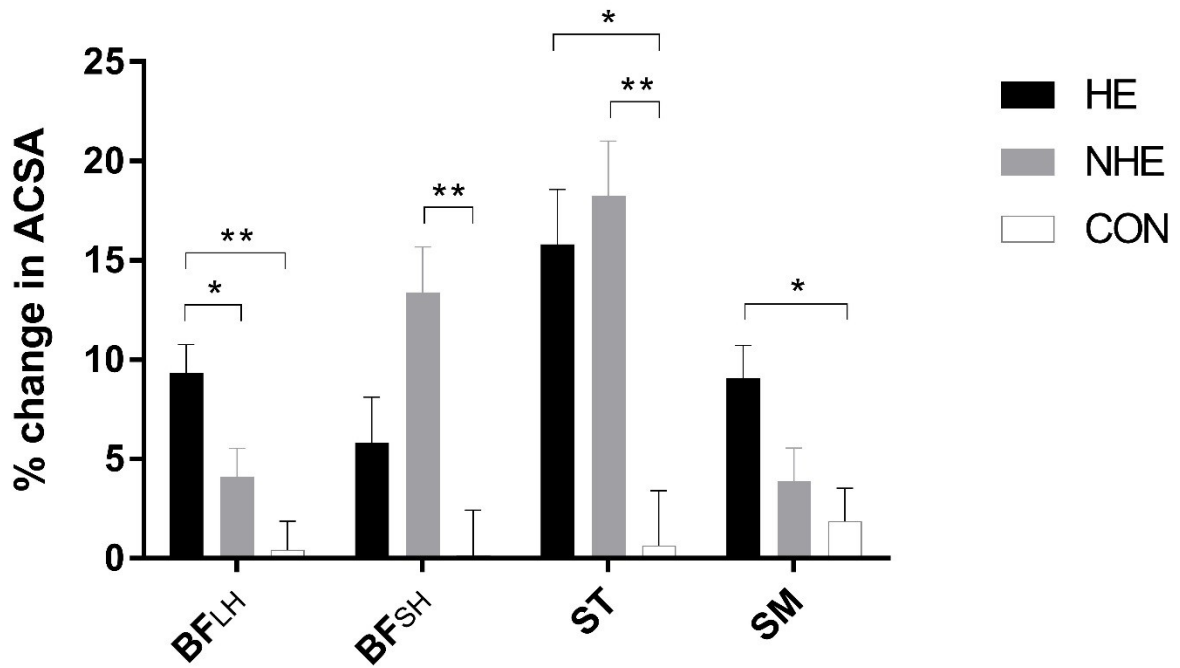
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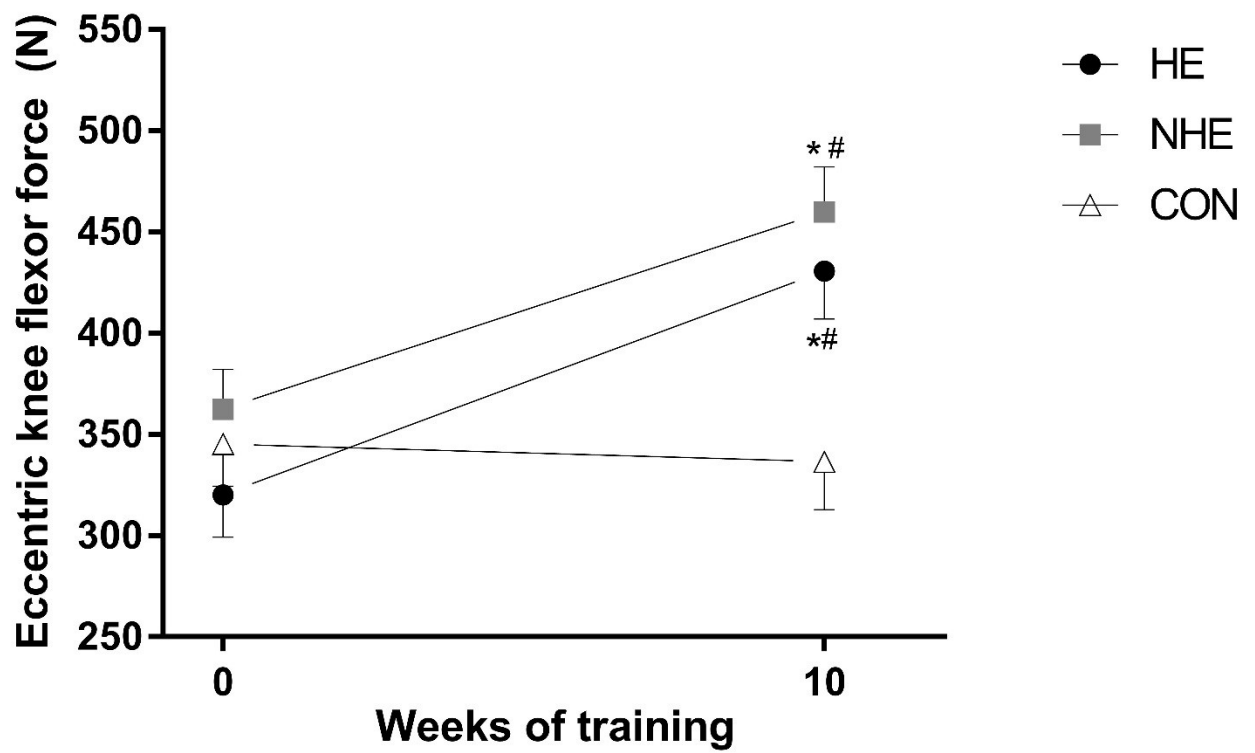
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